TOXICITY OF DDT RESIDUES:

Effect of Time of Exposure of Insects, Coverage and Tenacity

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CONTENTS

	4	PAGE
Introduction		3
Effect of Position of Exposure Cages		3
Uniformity of Spray Deposit		3
Uniformity Tests of Dosage Series		
Contamination of Cages		5
DISCUSSION OF VARIATION		6
The Effect of Coverage		7
EFFECT OF TIME OF EXPOSURE OF INSECTS		10
Relation Between Concentration and Time of Exposure of Insects		14
Effect of Tenacity		16
Methods		
Results		17
Effect of Method of Processing on Tenacity		29
Effect of Addition of a Wetting Agent		31
Effect of Addition of Fungicides		33
Summary		33
LITERATURE CITED		35

TOXICITY OF DDT RESIDUES:

Effect of Time of Exposure of Insects, Coverage and Tenacity

NEELY TURNER AND NANCY WOODRUFF

The large variation in results of tests of toxicity of DDT residues to house flies using certain laboratory methods has been reported by Woodruff and Turner (16). In these experiments, the residues were deposited on glass slides, to which house flies were attracted by light. After the flies were exposed for one to two hours, the treated slide was covered by a baffle to prevent further exposure. As a general rule, the deposit of DDT was varied by changing the concentration in the spray suspension or solution. Results varied considerably. During the studies reported (16), as well as those of Turner and Woodruff (15), constant attempts were made to isolate the factors responsible for the variations.

Effect of Position of Exposure Cages

In exposing the flies to the residues, it was necessary to stack the cages in front of the light source. Some were farther from the light source than others. In order to test the effect of the position of the cage on mortality of exposed flies, slides on twenty cages were sprayed with the same DDT mixture and exposed at the same time. There were the usual variations in mortality, but there seemed to be no correlation between the position of a cage in relation to the light source and mortality. A second test in which the same cages were used but in which their positions were switched showed even larger variations, but again there was no effect of position.

Uniformity of Spray Deposit

The deposits of DDT required to kill flies in the laboratory tests were too small to be weighed with accuracy. However, a series of slides sprayed for the same length of time with water showed a range of from .2187 g. to .2723 g. of deposit in one series of tests. Later tests after adjustment of the spray nozzle showed a range of from .1687 g. to .1873 g. of deposit, the coefficient of variation being 4.24 per cent. This did not seem large enough to account for the variations in mortality.

Uniformity tests were made by spraying 10 slides with the same concentration of DDT for the same length of time. House flies were exposed twice on successive days to each series of slides, and the mortality results have been summarized in Table 1. It is evident that there was a great deal of variation in mortality, too large to be explained by variations in deposit. For instance, the ninth slide in Test

TABLE 1. UNIFORMITY TESTS OF SERIES OF 10 SLIDES SPRAYED WITH THE SAME DDT

Each pair of tests represents two exposures of the same slides on successive days

				Per Cer	nt Morta	lity of H	ouse Flie	s		
	-	DDT	Concer	ntration	.0625%	1	DDT (Concenti	ration .0	312%
			Tes	st No.				Test N	lo.	
Slide No.	1	2	3	4	5	6	7	8	9	10
1	89.4	68.7	70.5	90.9	95.4	81.2	39.1	57.1	58.3	58.8
2	81.8	65.2	80.7	86.6	76.9	75.0	45.1	57.3	50.0	48.0
3	89.6	66.6	66.6	80.9	84.2	80.9	46.4	61.9	40.0	45.4
4	82.6	75.0	65.0	92.3	62.5	69.2	66.6	70.0	69.2	52.9
5	90.9	73.6	85.1	87.5	80.0	76.9	50.0	68.9	60.0	50.0
6	87.5	83.3	87.8	89.4	93.3	77.2	34.4	65.0	61.1	56.5
7	89.4	66.6	82.7	78.5	86.6	77.7	63.4	61.5	60.8	42.8
8	96.0	73.9	75.0	85.7	88.8	91.3	37.9	60.0	53.3	50.0
9	90.0	76.1	66.6	95.8	100.0	68.5	42.8	45.4	52.1	57.1
10	95.0	67.8	75.0	84.6	70.0	76.9	46.4	59.0	55.5	54.5

5 killed all of the flies while the tenth slide killed only 70 per cent. Test 6, in which the same slides were used, showed a higher kill, 76.9 per cent, for the tenth slide than for the ninth (68.5 per cent). Obviously, the low kill for slide 10 in Test 5 was not caused by a low deposit. In fact, of the 50 slides involved in the test, the only case in which there was any evidence of a low or a high deposit, as evidenced by successive low or high mortalities, was slide 4 in Tests 5 and 6.

Uniformity Tests of Dosage Series

A second type of uniformity test was made by spraying four slides in a dosage series and replicating four times in a single experiment. Results of two such tests are given in Table 2. Since the highest

Table 2. Uniformity Tests of Dosage Series of DDT Residues for Control of House Flies

Each pair of tests represents two exposures of the same slides

					•	
		Per C	ent Mortality	of House Flie	es	
Per Cent	Tes	t No.		Test	No.	
Conc. DDT	1	2	Mean	3	4	Mean
.125	100.0	100.0	100.0	100.0	100.0	100.0
.0625	91.4	88.0	89.7	57.9	100.0	78.9
.0312	85.2	73.9	79.5	76.5	95.0	85.8
.0156	23.8	57.1	40.0	42.9	73.7	58.1
.125	100.0	100.0	100.0	100.0	100.0	100.0
.0625	87.5	74.0	80.7	69.7	100.0	84.8
.0312	78.7	58.3	68.5	68.0	76.2	72.1
.0156	45.2	57.8	51.5	47.6	79.1	63.3
.125	100.0	100.0	100.0	100.0	100.0	100.0
.0625	92.3	82.6	87.9	82.2	76.5	79.3
.0312	73.6	65.7	69.6	71.4	53.3	62.3
.0156	33.3	54.2	43.7	56.0	36.8	46.4
.125	100.0	100.0	100.0	100.0	100.0	100.0
.0625	76.9	85.0	80.9	77.3	100.0	88.6
.0312	82.6	61.9	72.2	35.3	86.4	60.8
.0156	31.5	54.5	43.0	58.8	60.0	59.9

concentrations killed all of the flies in every test, only three mortalities could be plotted on logarithmic-probability paper. When curves were fitted to the points by inspection, the dosage in terms of concentration of DDT required to kill 80 per cent of the flies in the eight tests were as follows: .0312 per cent, .045, .037, .0625, .0625, .0625 and .045. The maximum variation was from .0312 to .0625 per cent (Figure 1). It is obvious that this method was capable of measuring differences greater than double in terms of dosage required for equal control, since the variation in the uniformity test was of this magnitude.

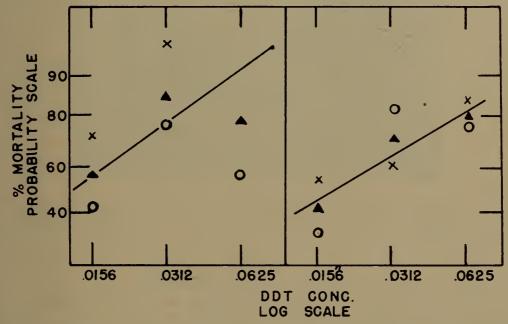


Figure 1. Extremes in mortality in dosage uniformity test. Highest kill at left, lowest at right. House flies exposed to DDT residues. The "scatter" of the points plotted is typical of the results with this type of test.

Contamination of Cages

Another possible source of variation was contamination of the fly cages. The type of exposure cage used in the tests (designed by Garman, 1943) had only one point of contact between the sprayed surface of the glass slide and the cage. Long use of the cages resulted in contamination of the slide holder. Such contamination occurred only after several months' use of the cages, and could be removed by washing with xylene or by application of a coat of shellac to the contaminated surface.

An inexpensive test cage which could be discarded if contaminated was designed and proved to be very satisfactory. This cage (Figure 2) was made of quarter-inch plywood of such a size that glass lantern slides could be used both as the sprayed surface and as a window. The glass plates were inserted in grooves made by a shallow sawcut in the plywood. Stiff paper masks were used to prevent the sprayed surface of the treated lantern slide from coming into contact with the wood of the cage. Stiff paper baffles were inserted in front of the mask at the end of the exposure period.



Inexpensive plywood exposure cage, showing sprayed slide in front, mask and baffle partly inserted. Flies and food are introduced through sliding glass panel at back.

DISCUSSION OF VARIATION

It is obvious that there is some serious source of variation in the method of testing DDT residues. Since the tests reported apparently show that the spray residues are reasonably uniform, the variation must be found either in the method of exposure or in the test insects. The attraction of the flies to the treated surface by means of light could be a source of variation. However, the method of Busvine and Barnes (2), in which the insects were forced to walk on treated surfaces, produced no more uniform results. House flies reared by the standard Peet-Grady technique have been assumed to be very uniform in resistance to chemicals. This is obviously not the case. On the other hand, Busvine and Barnes used a variety of test insects with results not substantially more uniform than with house flies. They reported, for instance, less mortality from 2 hours, 6 hours and 1 day exposure of bedbugs to DDT films than from 1 hour exposure.

Large variations in results are not confined to the residue method of testing. For instance, Lindquist and Wilson (10) reported large variations in the mortality of flies exposed to a fine mist spray containing DDT. There were also reversals in the order of susceptibility of ordinary flies and of special flies resistant to DDT, although on the average the special stock was much more resistant.

Busvine and Barnes (2) considered the problem of testing using direct spraying as compared with residues. Bliss (1) has shown that, if a constant amount of spray is applied to insects, the logarithm of

the concentration of the toxicant has a linear relationship to the mortality expressed in probits. Busvine and Barnes reasoned that for this relationship to hold in the case of residue testing, two assumptions must be made: "first, that the number of particles picked up by the insect is directly proportional to the number per unit area; second, that the toxic effect is related to the logarithm of the number of particles on the insect." The second assumption is the same whether spraying or residues are involved. However, as Busvine and Barnes pointed out, the accumulation of insecticides on the legs of insects walking over a treated surface is distinctly different from deposition of the same insecticides on the bodies of the same insects by spraying. The assumption that the number of particles picked up is directly proportional to the number per unit area seems entirely reasonable if the insects are forced to remain on the treated surface. This assumption might be questioned in the case of insects which could leave the treated area. Kennedy (9) has observed that mosquitoes seemed to be repelled by DDT deposits, and did not alight on them for as long a period as on untreated surfaces.

THE EFFECT OF COVERAGE

The method of using glass lantern slides allowed some types of test that should throw some light on the factors involved in variation. The exposed area was approximately 12 square inches. A series of tests on the relation between the size of area treated and mortality was carried out. The same amount of DDT was applied to each slide, but the area treated was varied. In all cases the treated area was centered on the slide, and areas of 1, 2, 4, 8 and 12 square inches were used. The results of two exposures for each of two sprayings are given in Table 3.

In this, as well as in most other tests described in this paper, the mortality of flies from the second exposure to any sprayed surface was usually lower than that from the first exposure. In other words, the deposit lost toxicity rather sharply. Part of this loss may be ascribed to actual removal of deposit by flies as they walk over the treated surface.

It will be seen that, in general, mortality was higher as the same amount of DDT was applied to a smaller area. In fact, the only important deviation was the highest concentration of the eight square inch series which averaged 91.2 per cent mortality. This appears to be a deviation and not a particular result of spraying eight square inches, since the three lower concentrations were in line with the results of the other tests. It is obvious, then, that the flies found the DDT on the one square inch as easily as they did when the entire slide was sprayed. It is also obvious that, within a square inch, they picked up the DDT somewhat in the order of the amount deposited on the surface. In fact, with the exception noted above, the points form logarithmic-probability curves reasonably well (Figure 3).

TABLE 3. EFFECT OF AREA SPRAYED ON MORTALITY OF HOUSE FLIES All slides have an equal amount of DDT deposited

	_		er Cent Mortali		
Area Treated	Test No.	Per	Cent Concentra	tion of DDT	
1 sq. in.		2.75	1.375	.688	.344
	1	100	100	67	70
	0	. 89	52	40	26
	2	· 76 65	100 46	78 48	59 48
	Mean	83	75	58	51
0 :	ivican				
2 sq. in.	-	1.375	.688	.344	.172
	1	100	92	73	75
	2	66 100	61 87	46 76	53 58
	4	64	33	36	37
	Mean	82	68	58	56
4 sq. in.		.688	.344	.172	.086
	1	93	94	55	74
		67	46	20	17
	2	96	96	73	45
		38	42	45	19
	Mean	74	69	48	39
8 sq. in.		.344	.172	.086	.043
	1	100	79	62	67
		100	40	10	50
	2	100 65	90 56	64 22	40 13
	Mean	91	66	39	42
12 sq. in.	1110411	.25	.125	.0625	.0312
12 sq. III.	-			•	
	1	96 48	83 16	56 14	47 0
	2	100	81	87 .	43
	_	42	25	50	43
	Mean	71	51	52	. 33

Horsfall (8) has shown that increasing the time of spraying and reducing the concentration of toxicant increased the effectiveness of fungicides in preventing the germination of fungous spores. He explained the results on the basis that coverage of a larger proportion of the area by increased spraying time left smaller unsprayed areas on which spores could germinate. In the case of the spores, no mobility of organism was involved. A spore fell at random on a sprayed or unsprayed area. House flies, being very active, might be expected to come into contact with DDT even though the entire surface was not treated.

Turner (14) found that small amounts of concentrated dusts were more effective in controlling Mexican bean beetles and cabbage worms than larger amounts of more dilute dusts. The reason, apparently,

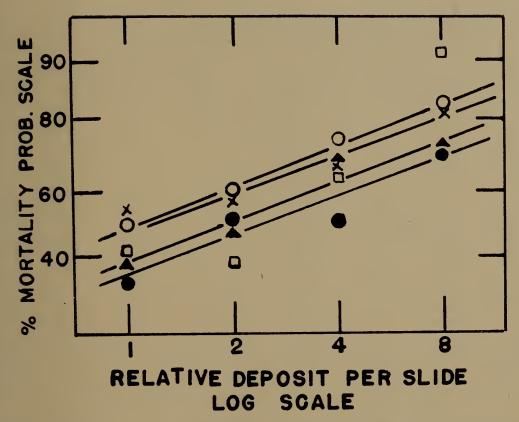


Figure 3. Effect of area treated on mortality. All tests had equal amount of DDT. o = 1 sq. in., x = 2 sq. in., a = 4 sq. in., a = 8 sq. in., and a = 12 sq. in. of exposed surface sprayed. Residue tests with house flies.

was that the probability of killing the insects with the dust that hit them was more important than the probability of hitting them at all. In the case of the residue tests with flies, the insects "found" the treated area, even if it was only one square inch, and were killed in somewhat larger numbers if the small treated area was treated with a high concentration of DDT. There were no indications of important deviations from parallelism among the dosage-response curves for the various areas (Figure 3). It is possible that the reduced toxicity of the less concentrated residues may have been caused by the excitant or repellent effect as noted by Kennedy (9) in tests on mosquitoes.

Parkin and Green (12) found that spraying for a short time with a concentrated solution of DDT produced a more toxic residue than the same amount of DDT per square foot applied as a more dilute solution. In their studies the total area sprayed was a constant.

One test of the relation between time of spraying, concentration and mortality was completed. In this test (Table 4), four slides were sprayed with each concentration at each spray time. Four exposures were made to determine which type of residue would lose effectiveness most rapidly. The results show no consistent differences among the spray times. Further, there was no appreciable difference in the rate of loss of effectiveness in the four spray times.

Table 4. Relation Between Concentration, Spray Time and Toxicity of Residues to House Flies

			Per Cent Mortality Exposure Number				
Per Cent							
Conc. DDT	Spray Time	1	2	3	4	Mean	
.25	2.5 secs.	100 80 80 95	79 98 100 67	83 54 73 71	67 26 73 45	74.4	
.125	5 secs.	92 .94 96 100	91 82 89 88	73 76 46 64	56 72 29 75	76.4	
.0625	10 secs.	93 50 76 63	100 92 94 81	97 73 82 83	42 20 65 67	73.6	
.0312	20 secs.	96 81 72 88	81 84 74 83	72 73 58 69	68 57 25 42	70,2	

EFFECT OF TIME OF EXPOSURE OF INSECTS

Busvine and Barnes (2) have considered the effect of time of exposure of insects to residues on toxicity. In general, they found no regular relationship between time of exposure and mortality with DDT as the toxicant. On the other hand, Parkin and Green (12) reported a definite increase in "knockdown" of flies as exposure time was increased.

Three series of tests involving time have been completed. In the first of these (Table 5), an exposure of 30 minutes produced a higher mortality than 15 minutes, 1 hour or 2 hours. In the second and third (Table 6) series, the mortality increased as the time of exposure was increased. It is obvious that exposure for two hours could not produce a lower mortality than any shorter period of time. Lower mortality would mean that if 100 insects were placed on a treated surface for 30 minutes, at which time 50 were removed, there would be a higher mortality than would result in the second group of 50 left on the surface for two hours. Such a result could mean only heterogeneity or an improper selection of the insects removed.

Table 5. Effect of Time of Exposure of House Flies to Residues on Mortality

			Per Cer	nt Mortality		
D C+	Time	-	Exposu	re Number		
Per Cent Conc. DDT	Exposed	1	2	3	4	Mean
		Test	1			
.25	15 min.	45	88	61	58	63
.125		79	56	30	10	44
.0625		86	26	25	15	38
.0312		27	33	46	9	29
.25	30 min.	83	90	100	100	94
.125		53	83	59	52	62
.0625		45	46	22	32	36
.0312		36	36	22	20	29
.25	1 hour	54	30	57	21	41
.125		92	43	63	53	63
.0625		14	22	44	37	29
.0312		42	30	6	4	21
.25	2 hours	92	52	100	61	76
.125		59	47	52	50	52
.0625		46	39	53	57	49
.0312		9	28	42	9	22
		Test	2			
.125 .0625 .0312 .0156	15 min.	87 67 56 38	52 29 40	100 75 48 30	46 26 30 10	78 55 41 29
.125	30 min.	94	73	100	100	92
.0625		47	46	76	36	51
.0312		56	21	74	53	51
.0156		39	24	47	26	34
.125	1 hour	100	50	100	100	88
.0625		74	42	100	62	69
.0312		59	48	71	52	57
.0156		49	45	52	19	41
.125	2 hours	100	67	100	95	90
.0625		94	58	96	81	82
.0312		71	48	95	50	66
.0156		69	46	70	30	54

TABLE 6. EFFECT OF TIME OF EXPOSURE OF

		Conce	entration DDT,	Per Cent by Wei	ight
Time Exposed	Test No.	2.75	1.375	.688	.344
	Sprayed	area one squa	re inch		
3.75 min.	1	86 93	17 47	40	31
	2	87	52	33 40	47 57
	3	71 62	37 48	26 35	42 23
	4	80 48	75 26	29 16	68 28
	Mean	75	43	31	42
7.5 min.	1	81	73	40	31
	2	89 68	72 58 59	73 69	38 54
	3	79 75	52	44 37	23 19
	4	80 35	80 23	64 36	29 12
	Mean	72	60	52	- 29
15 min.	1	67	80	29	36
15 min.	2	92 70	59 50	35 48	41 33
	3	69 95	15 56	45 38	17 60
	4	87 64	67 5	80 25	71 6
	Mean	78	47	44	41
30 min.	1	100	95 76	72	33
	2	90 91	76 90	39 76	24 81
	3	63 100	57 55	19 31	14 18
	4	83 79	87 29	72 39	65 11
	Mean	87	70	50	35
60 min.	1	100	92	87 38	83 50
	2	100 100	92 93 88 37 89 73 63	59	36
	3	86 95	89 70	23 50 -	27 40
	4	93 87	63	85 0	62 11
	Mean	96	76	49	44

House Flies to Residues on Mortality

	m .	Conce	entration DDT,	Per Cent by We	ight
Time Exposed	Test No.	1.375	.688	.344	.172
	Sprayed o	area two squar	e inches		
3.75 min.	1	21	0	4	5
	2	23 39	29 50	38 25	19 50
		43	45	53	61
	Mean	31	31	30	34
7.5 min.	1	30	6	_ 0	10
	2	63	25 30	29 58	20 48
		64	33	37	31
	Mean	52	24	31	27
15 min.	1	46	45	13	35
	2	90 78	87 48	33 42	17 33
		33	41	22	23
	Mean	62	56	28	27
30 min.	1	62	33	62	15
	2	100 89	88 79	70 73	25 18
	_	. 64	33	13	29
	Mean	79	58	54	22
60 min.	1	81	96	83	15
	2	95 91	96 86	88 71	81 44
	-	45	52	50	45
	Mean	78	83	73	46

On account of the variation in results, it is difficult to arrive at the mathematical relationship between time of exposure and mortality. The data seem to indicate, however, that the effect of time of exposure may be logarithmic (Figure 4). It is certainly not arithmetic. Ob-

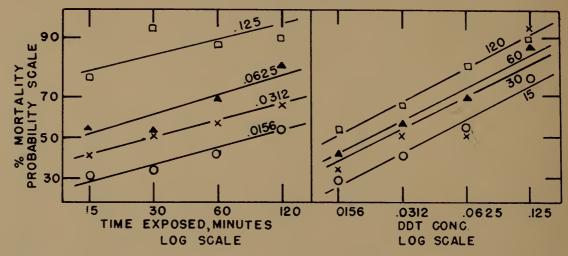


Figure 4. At left, effect of time of exposure on mortality at four different concentrations of DDT (Table 5, Test 2). At right, effect of concentration at four times of exposure in minutes from the same data.

viously, increasing the time of exposure allows the insects to pick up more toxicant and, therefore, acts in somewhat the same way as increased spray time or increased concentration. When the insects are not confined to the treated surface, but are allowed free access to it, increased time may mean increased probability of the insects walking over or alighting on the treated surface.

Relation Between Concentration and Time of Exposure of Insects

In the tests involving exposure time, four concentrations of DDT were used for each time of exposure. In most of the tests, the mortality increased with equal rapidity following increased time of exposure at all concentrations. In Table 5, Test 1, there is some indication that mortality increases more rapidly when the concentrations are increased, although the variation is large. In Test 2, this did not occur (Figure 4). In both tests reported in Table 6, the effect of time of exposure on mortality was much more marked as the concentration was increased (Figure 5). This indicates that there was an interaction between concentration and time of exposure, especially when the area was one or two square inches. Such an interaction might be explained by the repellent or excitant effect of DDT residues as noted by Kennedy (9). He emphasized the necessity of using higher concentrations to produce heavier residues and thus kill the mosquitoes before repelling them. MacInnes (11) has also commented on the "restlessness" of mosquitoes in treated cages.

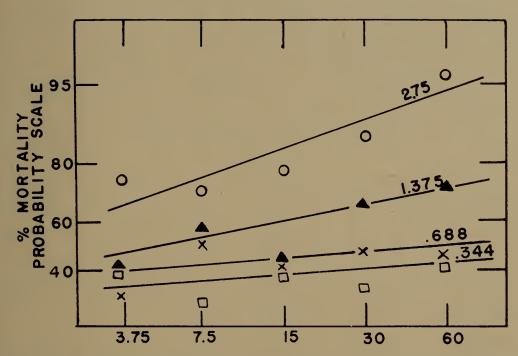
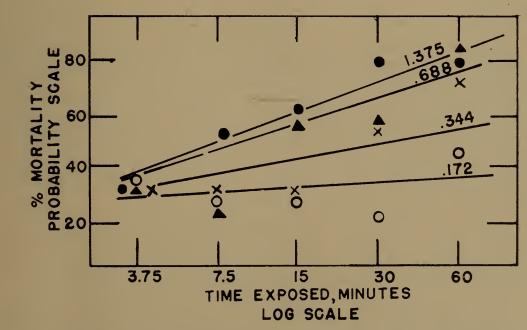


Figure 5A. Effect of time of exposure on four concentrations of DDT applied to one square inch of the surface.



B. Effect of time of exposure on four concentrations of DDT applied to two square inches of the surface.

EFFECT OF TENACITY

Spray powders of DDT used as water suspensions are usually made from equal quantities of technical DDT and a diluent. Chisholm (3) has pointed out that technical DDT softens at temperatures considerably below 88° C., making the addition of a dry diluent necessary for processing. Such a diluent may be expected to affect the toxicity of spray residues of such powders. Woodruff and Turner (16) have shown that such diluents may affect the particle size of the DDT which, in turn, affects its toxicity. However, factors other than particle size also influenced the toxicity of the products. The effect of diluents on tenacity is one of the obvious and important factors to be considered.

Garman (5) has studied the effect of "stickers" added to arsenate of lead. He found that skimmed milk powder, flour and soap did not improve the tenacity of lead arsenate, and actually reduced it when used in quantities of 25 per cent of the solid material in the spray. Oils, bentonite plus skimmed milk or casein, and aluminum gels, including bentonite, improved tenacity. With some materials there was a relation between the amount of material added and tenacity, but the good stickers never caused an actual reduction in the tenacity of lead arsenate when used in equal quantities.

Horsfall (8) has discussed the tenacity of fungicides and has pointed out that smaller particle size should increase tenacity, and that large deposits weathered more rapidly than small ones, a fact also noted by Garman (5). The effect of rainfall was found by Horsfall to be logarithmic, other things being equal.

Methods

Tenacity of diluents and of diluents with DDT was determined by a modification of the Heuberger method (7). Clean glass lantern slides were weighed, sprayed with the suspension by means of a precision laboratory sprayer, and weighed after drying to determine deposit. A series of slides having deposits of similar weight was swished through a large pan of water, using a series of 2, 4, 8, 16 and 32 strokes per slide. Since the size of the deposit is known to affect tenacity, four sizes of deposit were obtained by using four concentrations of each spray mixture. This design should provide a measure of any interaction between amount of deposit and amount of washing.

The diluents used were a series of samples of diatomaceous earths¹, a fuller's earth (*Attaclay*), and a pyrophyllite (*Pyrax ABB*). The pyrophyllite was used, not because it is preferred as a processing agent with DDT, but because of the type of tenacity it displays.

Deposits of DDT spray powders were tested for toxicity to house flies. Amounts small enough to kill less than 100 per cent of the flies were too small to be weighed with accuracy; therefore, only biological data were obtained for these mixtures.

¹Prepared by the Johns-Manville Research Laboratory, Manville, N. J.

Results

Deposition of the materials in relation to concentration in the suspension is given in Table 7. It will be noted that the diatomaceous earths deposited very much less material in relation to concentration than did the rest of the series. No reason can be given for this fact, but it is probably connected with the surface tension of the suspensions and its effect on deposition. [See Turner and Woodruff (15)].

Table 7. Relation Between Concentration of Material in Sprays and Average Size of Deposit Per 13 Square Inches

Material	Per Cent Concentration	Av. Deposit—Gram
54	.125 .25 .5 1.0	.0004 .0007 .0013 .0021
61	.125 .25 .5 1.0	.0003 .0006 .0014 .0026
62	.125 .25 .5 1.0	.0005 .0008 .0012 .0026
64	.0625 .125 .25 5	.0004 .0006 .0009 .0012
67	.125 .25 .5 1.0	.0007 ¹ .0011 .0015 .0021
69	.125 .25 .5 1.0	.0010 ¹ .0013 .0016 .0020
fuller's earth	.125 .25 .5 1.0	.0008 ¹ .0014 .0029 .0053
pyrophyllite	.125 .25 .5 1.0	.0009 .0016 .0024 .0052

¹Two replicates.

A summary of the results of the effect of washing on residues is given in Table 8. Dosage-response curves were fitted to these data plotted on logarithmic-probability paper and the number of strokes required to remove 50 per cent of the deposit calculated in Table 9. Several points appear of immediate interest. Pyrophyllite and fuller's earth were of comparatively low tenacity. Diatomaceous earth 67 was the most tenacious material. Size of deposit had relatively little effect on tenacity of fuller's earth and pyrophyllite. Among the other materials, diatomaceous earth 69 followed the pattern of decrease in

TABLE 8. TENACITY OF DILUENTS SPRAYED ON GLASS SLIDES IN SUSPENSION

			Per Cent Te	nacity	
Material	No. Strokes Washed		Av. Size De	posit	
		.0004	.0006	.0009	.0012
64	2	33.3	33.3	66.7	92.4
	4	66.7 40.0	75.0 66.7	77.8	75.0 75.0
	2 4 8 16 32	60,0	50.0	20.0	45.5
	32	25.0	33.3	33.3	58.4
		.0004	.0007	.0013	.0021
54	2	60.0	57.2	46.2 57.2	77.3
	4 8	20.0 25.0	42.9 37.5	57.2 41.7	68.2 62.0
	2 4 8 16 32	20.0	28.6	30.8	52.7
	32	0	25.0	53.9	45.5
		.0010	.0013	.0016	.0019
69	2 4 8	55.6 54.6	41.7 53.9	37.5 37.5	50.0 42.2
	8	44.5	35.8	35.3	36.9
	16 32	20.0 18.2	30.8 16.7	13.4 20.0	30.0 27.4
	34	.0003	.0006	.0014	.0026
61	2	50.0	33.3	35.8	64.0
01	4	66.7	50.0	21.5	60.0
	8 16	25.0 66.7	42.9 28.6	26.7 33.3	56.0 46.5
	8 16 32	50.0	50.0	50.0	40.8
	- College	.0005	.0008	.0012	.0026
62	2	60.0	50.0	77.8	92.4
	4 8	40.0 40.0	66.7 57.2	46.2 37.5	73.1 79.2
	8 16	20.0	42.9	68.9	61.6
	32	20.0 .0007	57.9 .0011	50.0 .0015	79.4
-					
67	2 4	50.0 57.2	72.8 45.5	53.4 31.3	85.8 60.0
	8	42.9	58.4	46.7	68.2
	16 32	71.5 37.5	50.0 63.7	14.3 64.3	61.2 62.0
	02	.0007	.0015	.0029	.0050
fuller's earth	2	37.5	20.0	34.5	25.5
idioi b odi oii	4	14.3	20.0	28.6	24.0
	4 8 16	0	13.3 13.3	26.7 20.7	20.9 12.8
	32	ŏ	13.3	17.3	18.8
		.0011	.0021	.0031	.0059
pyrophyllite	2	41.7	50.0	59.4	51.7
	2 4 8	20.0 17.9	52.7 42.9	40.0 37.5	39.0 27.1
	16	7.3	40.9	20.0	26.7
	32	0	26.4	16.1	25.0

Material	Deposit	No. Strokes to Remove 50%	Material	Deposit	No. Strokes to Remove 50%
64	.0004 .0006 .0009 .0012	2 6 12 32	62	.0004 .0008 .0012 .0026	3 6 10 7,0
54	.0004 .0007 .0013 .0021	1 3 7 20	67	.0004 .0007 .0017 .0035	1 2 100 >1000
69	.0011 .0018 .0029 .0054	16 6 3 1	fuller's earth	.0008 .0014 .0029 .0053	$ \begin{array}{c} 3^{1} \\ 10^{1} \\ 18^{1} \\ 3^{1} \end{array} $
61	.0003 .0006 .0014 .0026	1 8 1 10	pyrophyllite	.0009 .0016 .0024 .0052	2 6 3 2

TABLE 9. TENACITY OF DILUENTS IN RELATION TO SIZE OF DEPOSIT

tenacity as the deposit was increased, as noticed by Horsfall (8) and Garman (5). The other five materials reversed this trend, and the heavier deposits showed a higher tenacity.

A single layer of particles on a surface would be affected only by the tenacity of the particles for the surface. This would be most nearly represented by the lowest deposits. As layers of particles of the same material are added, the tenacity of the particles for others of the same kind is involved. Thus, size of deposit may have had no effect on tenacity of fuller's earth and pyrophyllite because the particles had low tenacity for each other and for the glass surface. Diatomaceous earth 69 may have less tenacity between particles than for the surface; therefore, low deposits had relatively better tenacity. The remaining materials stuck to each other better than to the sprayed surface; hence, tenacity increased with increased deposit.

Of this series of materials, fuller's earth, pyrophyllite, and diatomaceous earths 67 and 69 were selected for further study, and for tests of tenacity as diluents for DDT wettable powders. Further tenacity tests were completed to provide data for dependable dosage-response curves (Table 10 and Figure 6). When the data for different deposits were plotted, the family of curves for pyrophyllite and diatomaceous earth 67 appeared to diverge, which indicates increasing differences between deposits as the amount of weathering was increased. The curves for diatomaceous earth 69 converged, indicating that heavy washing had about equal effect on all sizes of deposit. For fuller's earth, three of the four curves for size of deposit converged, while the curve for the lowest deposit was very much steeper (the data showed no tenacity at all at 32 strokes).

Although there seems to be no way to apply pure DDT of crystal size and shape similar to that of spray powders, it can be assumed that DDT itself has a high tenacity against rainfall. For instance, Gunther et al (6) found that DDT deposits applied in kerosene to the bark of trees decreased at a relatively uniform rate, regardless of rainfall. Hors-

Strokes to remove 70%.

TABLE 10. TENACITY OF DILUENTS SPRAYED

Material	No. Strokes Washed			Per Cent Te	nacity	
Material	W astred		.0011	.0018	.0029	.0054
69	2		66.7 72.8	47.6 60.0	43.8 59.3	36.5 53.9
		mean	69.8	53.8	51.6	45.2
	4		50.0 81.8	42.9 58.9	44.9 51.7	37.8 47.1
		mean	65.9	50.9	48.3	42.6
	8		38.5 63.7	52.9 46.7	40.0 46.7	36.5 39.6
		mean	51.1	49.8	43.4	38.1
	16		40.0 55.6	31.6 54.6	31.3 48.0	32.1 40.0
		mean	47.8	43.1	39.7	36.1
	32	,	36.4 40.0	23.9 50.0	29.6 35.8	27.8 34.0
		mean	38.2	37.0	32.7	30.9
			.0004	.0007	.0017	.0035
67	2		42.9 28.6	55.6 40.0	83.4 57.7	77.5 75.6
		mean	35.8	47.8	70.1	76.1
	4		33.3 14.3	62.5 41.7	78.3 60.9	81.6 76.9
		mean	23.9	52.1	69.6	79.3
	8	,	33.3 16.7	33.3 30.0	65.0 60.9	75.8 83.3
		mean	25.1	31.7	63.0	79.6
	16		14.3 14.3	41.7 30.0	60.0 55.0	76.2 67.4
		mean	14.3	36.9	57.5	71.3
	32		14.3 0	37.3 50.0	57.2 55.5	63.9 77.3
		mean	8.2	38.7	56.1	70.6

ON GLASS SLIDES IN SUSPENSION

Material	No. Strokes Washed		 	Per Cent Ten Av. Size Der		
			 .0009	.0016	.0024	.0052
pyrophyllite	2		41.7 50.0	50.0 60.0	59.4 68.8	51.7 52.3
		mean	45,9	55.0	64.1	52.0
	4		20.0 37.5	52.7 66.7	40.0 40.0	39.0 45.5
		mean	28.8	59.7	40.0	42.3
	8		17.9 50.0	42.9 60.0	37.5 41.2	27.1 34.1
		mean	34.0	57.5	39.4	30.6
	16		7.7 25.0	40.9 35.7	20.0 40.0	26.7 27.1
		mean	16.4	38.3	30.0	26.9
	32		$\begin{matrix} 0 \\ 12.5 \end{matrix}$	26.4 38.5	16.1 33.3	25.0 25.0
		mean	6.3	32.5	24.8	25.0
			 .0008	.0014	.0029	.0053
fuller's eartl	h 2		50.0 37.5	61.5 20.0	50.0 34.5	38.6 25.5
		mean	43.8	40.8	42.3	32.1
	4		33.3 14.3	58.5 20.0	48.2 28.6	35.8 24.0
		mean	23.9	39.2	38.4	29.9
	8		20.0 0	42.9 13.3	39.3 26.7	34.0 20.9
		mean	10.0	28.1	33.3	27.5
	16		10.0 0	40.0 13.3	30.0 20.7	32.1 12.8
		mean	5.0	26.7	35.4	22.5
	32		0	33.3 13.3	28.6 17.3	25.4 18.8
		mean	0	23.4	23.0	22.1

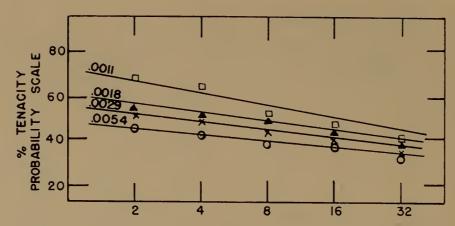
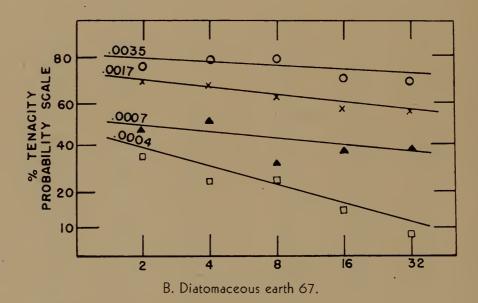
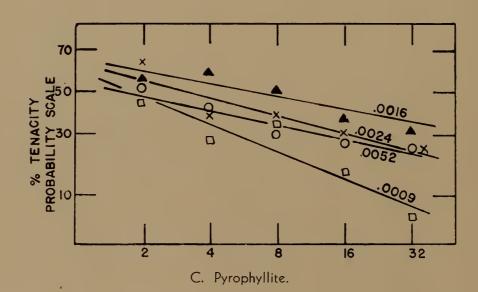


Figure 6. Effect of amount of washing on four sizes of deposit of:
A. Diatomaceous earth 69.





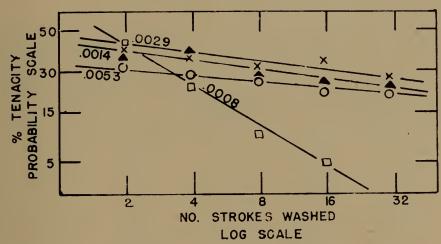


Figure 6. D. Fuiler's earth.

fall (8) has shown that diluents reduce the tenacity of red copper oxide, which has greater tenacity than the diluents used with it. In the case of DDT, there are the following possibilities: (1) a diluent of high tenacity might remain with the deposit and actually cover up the DDT so that toxicity of the residues would be relatively lower than toxicity of undiluted DDT; (2) a diluent of low tenacity might be removed from the deposit, leaving the DDT exposed and possibly resulting in increased toxicity after weathering as compared with before; (3) a diluent of low tenacity might wash off readily, removing the DDT with it. In alternate (1) the toxicity of residues would be relatively unchanged by washing, in (2) toxicity would tend to increase on light washing, and in (3) toxicity would decline steadily with increased washing.

Before presenting the data on toxicity of DDT spray powders, it would be well to point out that total deposits were lower than the deposits of pure diluents discussed above. If extrapolation is possible, diatomaceous earth 69 might be expected to be the most tenacious, followed by diatomaceous earth 67, pyrophyllite and fuller's earth. Fuller's earth might be expected to weather most rapidly as amount of washing increased. Further, because of the interactions involved, simple rectilinear dosage-response curves might not be expected.

Preliminary tests of the effect of washing on spray powders prepared from materials listed in Table 8 were made by testing the toxicity of the residues to house flies. The results have been summarized in Table 11. In general, these results show (1) relatively little loss in toxicity after as much washing as 32 strokes, which is a rather drastic test, and (2) relatively little effect of the diluent on toxicity after washing. Pyrophyllite used alone, for instance, had very low tenacity, but on the whole the washing did not reduce the toxicity of pyrophyllite-DDT mixtures in any consistent manner. Fuller's earth alone showed very little tenacity, but in no case did washing reduce the toxicity of fuller's earth-DDT mixture drastically. Small deposits of diatomaceous earths 54, 61, 62, 64 and 67 had low tenacity. When used with DDT and compared with unwashed deposits, all but material 64 lost toxicity rather consistently, especially at the higher concentrations. Sample 69 with DDT lost toxicity only at one concentration.

TABLE 11. TOXICITY OF RESIDUES OF DDT SPRAY POWDERS Laboratory tests

			Per Cent Mortality			
Diluent	Per Cent Conc. DDT	No. Strokes Washed	1	2	Average	
64	.0475	0 4 8 16 32	24.0 44.4 22.4 45.8 38.2	66.9 42.7 38.7 22.9 42.8	45.4 43.5 30.5 34.5 40.5	
	.095	0 4 8 16 32	53.6 65.4 39.2 51.3 32.2	63.2 55.6 62.7 44.8 40.0	58.4 65.5 50.9 48.0 36.1	
	.19	0 4 8 16 32	59.9 76.6 77.7 89.1 68.0	75.0 59.7 76.6 52.1 74.7	67.4 68.1 77.6 70.6 71.3	
	.38	0 4 8 16 32	61.4 95.0 100.0 89.2 80.1	96.4 100.0 83.0 97.6 77.1	78.9 97.5 91.5 93.4 78.6	
61	.0475	0 4 8 16 32	64.7 60.8 59.5 51.5 100.0	2.7 21.3 2.1 10.4 3.4	33.7 41.0 30.8 30.9 51.7	
	.095	0 4 8 16 32	52.9 28.5 32.8 54.3 36.8	36.5 11.6 90.1 10.0 0	44.7 20.0 61.4 32.1 18.4	
	.19	0 4 8 16 32	76.0 54.4 57.8 52.7 65.6	52.2 45.4 28.7 30.5 19.2	64.1 49.9 43.2 41.6 42.4	
	.38	0 4 8 16 32	95.4 60.7 70.0 53.3 82.5	66.4 35.4 52.0 47.9 44.1	80.9 48.0 61.0 50.6 63.3	

MADE FROM DIFFERENT DILUENTS AS AFFECTED BY TENACITY on house flies

	Pon Cont	No. Strokes	I	Per Cent Mortality		
Diluent	Per Cent Conc. DDT	Washed	1	2	· Average	
54	.0312	0 4 8 16 32	41.5 49.1 9.4 41.6 24.2	56.2 64.8 9.6 11.1 50.5	48.8 56.9 9.5 26.3 37.3	
	.0625	0 4 8 16 32	67.6 41.6 58.9 53.3 16.1	15.2 - 45.9 100.0 20.6 28.5	41.4 43.7 79.4 36.9 22.3	
	.125	0 4 8 16 32	100.0 77.0 66.1 45.4 55.0	69.4 34.9 30.5 68.7 60.4	84.7 55.9 48.3 57.0 57.7	
	.25	0 4 8 16 32	88.3 93.4 100.0 83.3 65.9	66.6 75.7 48.9 35.3 62.5	77.4 84.5 74.4 59.3 64.2	
69	.0312	0 4 8 16 32	37.7 40.0 59.5 49.7 47.2	48.4 40.8 34.1 26.6 42.5	43.0 46.8 46.8 38.1 44.8	
	.0625	0 4 8 16 32	51.7 63.3 67.6 45.3 61.5	36.5 82.0 50.0 54.3 61.2	44.1 72.6 58.8 49.8 61.3	
	.125	0 4 8 16 32	85.2 70.8 73.5 60.4 65.0	85.0 69.7 79.5 79.1 68.0	85.1 70.2 76.5 69.7 66.8	
	.25	0 4 8 16 32	66.4 68.1 67.5 61.8 81.5	82.4 93.1 74.4 64.6 93.0	74.4 80.6 70.9 63.1 87.2	

TABLE 11 (CONTINUED). TOXICITY OF RESIDUES OF DDT SPRAY Laboratory tests

	Por Cont	No Stroke	Per Cent Mortality		
Diluent	Per Cent Conc. DDT	No. Strokes Washed	1	2	Average
62	.0312	0 4 8 •16 32	27.7 2.1 0 15.4 0	30.1 29.6 31.9 17.0 24.5	28.9 15.8 15.9 16.2 12.2
	.0625	0 4 8 16 32	31.0 32.9 5.2 20.0 19.2	23.4 20.2 39.1 20.8 • 15.0	27.3 26.5 22.1 20.8 17.1
	.125 '	0 4 8 16 32	100.0 31.4 36.4 40.0 45.6	58.6 56.9 17.2 37.7 14.9	79.3 44.1 26.8 38.8 30.2
	.25	0 4 8 16 32	42.5 50.1 65.7 30.3 51.8	57.0 25.0 30.9 16.4 28.0	49.7 37.5 48.3 23.3 39.9
fuller's	s earth .0475	0 4 8 16 32	24.9 42.8 34.3 2.0 11.9	43.1 40.2 30.0 38.2 31.2	34.0 41.5 32.1 20.1 21.5
	.095	0 4 8 16 32	25.0 16.2 20.8 5.0 50.1	33.1 30.3 63.3 47.3 66.6	29.0 23.2 42.0 26.1 58.3
	.19	0 4 8 16 32	41.4 35.1 27.7 43.3 100.0	54.5 62.6 40.0 49.9 57.6	47.9 48.8 33.8 46.6 78.8
	.38	0 4 8 16 32	46.5 88.6 56.0 53.8 75.9	84.6 71.6 72.1 48.8 59.2	65.5 80.1 64.0 51.4 67.5

POWDERS MADE FROM DIFFERENT DILUENTS AS AFFECTED BY TENACITY on house flies

	Por Cont	No. Strokes	Per Cent Mortality			
Diluent	Per Cent Conc. DDT	Washed	1	2	Average	
67	.0312	0 4 8 16 32	37.5 34.2 23.5 26.9 50.4	14.5 22.0 7.6 40.1 0	26.0 28.1 15.5 32.5 25.2	
	.0625	0 4 8 16 32	49.4 42.3 13.1 39.5 44.6	26.6 29.1 35.2 46.6 40.8	38.0 35.7 24.1 42.5 42.7	
	.125	0 4 8 16 32	83.3 18.7 52.6 34.3 38.9	69.7 40.9 27.7 57.5 51.7	76.5 29.8 40.1 45.9 45.3	
	.25	0 4 8 16 32	78.5 45.1 53.3 20.3 53.7	88.4 66.6 65.9 52.6 100.0	83.4 55.8 59.6 36.4 76.8	
pyroph	yllite .125	0 1 2 4 8 16	11.0 0 16 16 30 38			
	.25	0 1 2 4 8 16	8 27 18 24 9 0			
	.5	0 1 2 4 8 16	9 21 5 22 0 6			
	1.0	0 1 2 4 8 16	38 40 33 21 19 22			

There were occasional cases of apparently increased toxicity following washing. However, these occurred in small numbers except with diluents 69 and 64.

These preliminary tests seemed to show that the toxicity of DDT in spray powders was not affected very seriously by the lack of tenacity of the diluent. Furthermore, the principal interaction between size of deposit and tenacity seemed to be lower tenacity of the higher deposits. In order to study the effects more fully three materials were selected, fuller's earth and diatomaceous earths 67 and 69. The results are given in Table 12 and show that, in general, diatomaceous earth 69-DDT lost less toxicity after washing than did material 67-DDT, as was to be expected from the results of tests of pure diluents at low deposits. Likewise, sample 69-DDT proved to be more tenacious than fuller's earth-

Table 12. Toxicity of Residues of DDT Spray Powders Made From Different Diluents as Affected by Tenacity

Laboratory tests on house flies

			Per Cent Mortality No. Strokes Washed					
	D 0 .	m						
Diluent	Per Cent Conc. DDT	$egin{array}{c} \operatorname{Test} \ \operatorname{No}. \end{array}$	0	4	8	16	32	
fuller's	earth .125	1	85	46	77	51	45	
			75	26	60	32	56	
		2	68	65	78	75	44	
			59	19	64	38	9	
		3	89	79	67	35	35	
			93	83	7	45	35	
		4	96	70	73	76	46	
			67	50	13	62	27	
		5	86	65	75	55	52	
			85	71	65	60	35	
		6	48	29	73 -	78	53	
			60	73	60	38	52 35 53 32	
		Mean	76	63	59	54	39	
69	.125	1	94	67	73	67	81	
			60	23	4	74	49	
		2	92	74	76	42	53	
			86	29	39	20	17	
		3	100	71	71	38	74	
			80	75	50	32	50	
		4	100	93	88	96	89 50	
			92`	75	50	32	50	
		5	100	88	100	80	87	
			100	85	82	73	58	
		6	76	100	100	95	82	
			96	61	67	57	50	
		Mean	90	70	67	67	62	
67	.125	1	57	94	65	81	42	
			71	50	62	34	46	
		2	88	74	78	79	71	
			64	65	40	50	43	
		3	79	45	53	20	54	
			88	67	66	53	100	
		Mean	75	66	61	53	59	

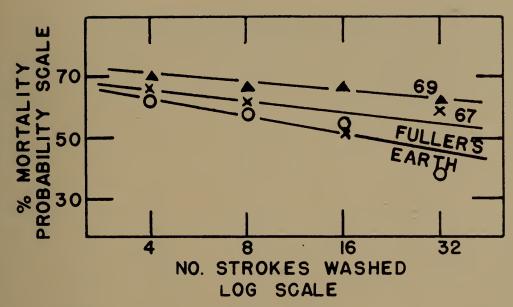


Figure 7. Effect of degree of washing on toxicity of residues to house flies of DDT spray powders processed from three diluents.

DDT, although the differences were not great. In terms of slope of the curves plotted from the data (Figure 7), those for diatomaceous earths 67 and 69 appeared to be parallel. There was a slight tendency for a steeper slope for fuller's earth in comparison with diluent 69.

It seems obvious that the materials of low tenacity did not wash off and remove DDT in the process, to any great extent. On the other hand, there was some evidence that they washed off and left the DDT on the surface. The superior tenacity of diatomaceous earth 69 at low deposits was apparently reflected in the relatively high toxicity of washed residues to house flies.

EFFECT OF METHOD OF PROCESSING ON TENACITY

Woodruff and Turner (16) have shown that the method of processing has an effect on the particle size and toxicity of DDT in spray powders. Mixtures prepared by pulverizing mechanical mixtures of DDT and the diluent were less effective than those made by dissolving the DDT in a volatile solvent, mixing the solution with the diluent, drying and micropulverizing, because the latter method usually produced smaller particle size. The impregnation method might be expected to have an effect on tenacity because it should provide a more intimate mixture of the DDT and diluent.

The effect of preparation by impregnation was studied and summarized in Table 13. These data may be compared with the data for the same diluents in Table 11. They show that the impregnation process had little effect on relative tenacity of 69 and 67 mixtures. With fuller's earth, there was some evidence that impregnation reduced the tenacity (Figure 8). This could be explained by the relatively poor tenacity of fuller's earth. Impregnation would allow the fuller's earth to remove more DDT than if a mechanical mixture were used.

Table 13. Effect of Impregnating Spray Powders With DDT on Tenacity and Toxicity of Residues to House Flies

	Par Cant	No Strokes	Per Cent Mortality			
Diluent	Per Cent Conc. DDT	No. Strokes Washed	1	2	Average	
69	.0312	0 4 8 16 32	73 23 85 66 37	33 47 43 63 40	53 35 64 64 38	
	.0625	0 4 8 16 32	38 67 48 15 53	64 66 47 60 54	51 66 47 33 53	
	.125	0 4 8 16 32	83 83 66 84 76	57 64 79 43 63	70 74 73 63 70	
	.25	0 4 8 16 32	100 95 90 58 85	75 95 82 94 83	87 95 86 76 84	
67	.0312	0 4 8 16 32	43 59 40 54 63	63 15 47 35 32	53 37 44 45 47	
	.0625	0 4 8 16 32	58 62 52 81 72	26 52 84 57 57	42 57 68 69 64	
	.125	0 4 8 16 32	100 87 68 67 86	54 58 60 43 21	77 72 64 55 53	
	.25	0 4 8 16 32	97 98 94 80 94 -	78 81 75 67 80	88 89 84 74 87	
fuller's earth	.0312	0 4 8 16 32	48 58 59 59 55	37 48 14 51	48 47 54 37 53	
	.0625	0 4 8 16 32	57 62 77 75 66	42 32 54 58 36	49 47 66 66 51	

Table 13 (Continued). Effect of Impregnating Spray Powders With DDT on Tenacity and Toxicity of Residues to House Flies

	Dev Cont	N - C+l	Per Cent Mortality			
Diluent	Per Cent Conc. DDT	No. Strokes Washed	1	2	Average	
	.125	0	79	83	81	
		4	95	57	76	
		8	57	51	. 54	
		16	52	83	67	
		32	72	43	58	
	.25	0	100	96	98	
		4	91	82	86	
		8	75	82	78	
		16	68	95	81	
		32	52	84	68	

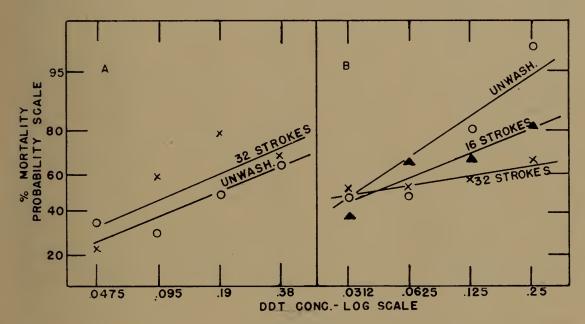


Figure 8. Effect of washing on toxicity of residues of DDT and fuller's earth. Mechanical mixture at left, impregnated mixture at right.

Effect of Addition of a Wetting Agent

Turner and Woodruff (15) showed that addition of a wetting agent to DDT spray powders resulted in a lower deposit of DDT on sprayed surfaces, but apparently did not reduce the toxicity of the residues. The effect of a wetting agent on tenacity was studied by adding $Triton X-100^{\circ}$ to the DDT spray powder made with diatomaceous earth 69 as a diluent. The amounts of deposit were too small to be weighed accurately; therefore, a test was made of toxicity of the residues to house flies (Table 14). Addition of Triton X-100 at increasing rates of from .125 to .5 per cent on the basis of the water in which the powder was suspended progressively reduced the toxicity of unwashed residues. This was to be expected if the amount of residue was reduced by use

¹Rohm and Haas Co., commercial product. Polyethylene glycol phenylisooctyl ether.

of the wetting agent. However, within each group of slides sprayed with the same amount of DDT and wetting agent, the washed slides were usually as toxic as the one not washed. In other words, the residue which was present was not reduced in tenacity by the use of the wetting agent.

TABLE 14. EFFECT OF WETTING AGENTS ON TENACITY OF RESIDUES OF DDT SPRAY **Powders** Toxicity tests on house flies

337-44*		No. Strokes	Per Cent Mortality		
Wetting Agent	Amount	No. Strokes Washed	1	2	Average
		Diluent 69			
None	None	0 4 8 16 32	92 91 82 85 86	96 80 62 50 55	94 85 72 70 70
Triton X-100	.125%1	0 4 8 16 32	59 70 77 33 71	76 79 73 48 71	67 74 75 40 71
,,	.25%1	0 4 8 16 32	43 28 17 55 47	46 22 67 65 31	44 25 42 60 39
,,	.5%1	0 4 8 16 32	22 52 63 23 73	50 19 76 50 15	36 35 69 36 44
		Diluent 67			
None	None	0 4 8 16 32	88 74 78 79 71	64 65 40 50 43	76 69 59 64 58
Vatsol K	1%1	0 4 8 16 32	82 92 65 69 67	60 56 60 69 30	71 74 63 69 48

¹On the basis of the water in which the powder was suspended.

Addition of $Vatsol K^1$ at the rate of 1 per cent of the water used reduced toxicity of unwashed residues slightly, but did not seem to reduce toxicity of washed residues to a substantially greater degree than in the test where no wetting agent was used.

¹American Cyanamid & Chemical Corp. 33 per cent sodium dioctyl sulfosuccinate and an inert filler.

Effect of Addition of Fungicides

Laboratory prepared Bordeaux mixture (4-2-50) and commercial copper oxide were added to DDT spray powder and tested for tenacity (Table 15). Bordeaux mixture appeared to reduce the toxicity of residues slightly on unwashed slides. Copper oxide apparently increased toxicity. With both materials, there was no evidence from toxicity of washed slides that the tenacity of the DDT had been affected adversely.

Table 15. Effect of Fungicides Added to DDT Spray Powders on Tenacity Toxicity tests on house flies

	NT - C+1		Per Cent Mortality		
Fungicide	No. Strokes Washed	1	2	Average	
None	0	. 89	95	92	
	4	100	94	97	
	8	100	100	100	
	16	100	100	100	
	32	95	100	97	
Bordeaux mixture					
(4-2-50)	0	80	64	72	
,	4	96	80	88	
	8	100	94	97	
	16	100	94	97	
	32	87	94	90	
Copper oxide 1.5%	0	100	100	100	
	$\overline{4}$	96	95	95	
	8	100	91	95	
	16	100	100	100	
	32	100	100	100	

Skaptason and Blodgett (13) found that cuprous oxide supplemented the control of potato pests by pyrethrum and in some cases by rotenone as well.

SUMMARY

Some of the factors which might cause variation in results of tests for toxicity of DDT residues using house flies have been studied. The position of the cages in front of the source of light was not involved. Variation in weight of deposit was not large enough to account for variation in results, and uniformity tests showed little evidence of serious variation in deposit. Contamination of test cages with DDT was a factor, but could be avoided by washing the cages with xylene, or by use of a new type of inexpensive plywood cage. The variation was not confined to house flies, since Busvine and Barnes (2) reported similar variation with other test insects.

Application of the same amount of DDT in each cage to areas of 1, 2, 4, 8 and 12 square inches resulted in a higher mortality when the DDT was applied to the smaller areas. When the coverage was changed by using a smaller concentration of DDT in the suspension and spraying for a longer time, no important differences in mortality of house flies resulted.

The effect of time of exposure was apparently logarithmic. There appeared to be an interaction between concentration and time of exposure when one or two square inches were treated. Dosage-response curves for higher concentrations were steeper than those for lower concentrations. This may be caused by the repellent effect of DDT as noted by Kennedy (9).

A study of some diluents which may be used in DDT spray powders showed that the diluents themselves added to water and sprayed on surfaces varied in amount of residue deposited. Fuller's earth and pyrophyllite deposited larger residues than the various diatomaceous earths. Tests of tenacity of residues of the diluents sprayed on glass slides showed that pyrophyllite and fuller's earth had relatively low tenacity and tenacity did not seem to be affected by size of deposit. Five of the diatomaceous diluents had increased tenacity as the size of deposit was increased. This may have been caused by the high tenacity of particles of these materials for other particles of the same material.

Further study of four diluents showed that diatomaceous earth 67 had the greatest tenacity, and pyrophyllite and fuller's earth the least. Diatomaceous earth 69 was intermediate, but had an advantage in that heavy washing seemed to affect all sizes of deposit equally. Toxicity tests of washed residues of these diluents processed with DDT to form spray powders showed surprisingly little difference. The diluents of low tenacity did not wash off and remove DDT. The superior tenacity of small deposits of diatomaceous earth 69 showed to some advantage.

Preparation of spray powders by the method of impregnation had little effect on tenacity, except that the product made with fuller's earth seemed to lose more toxicity after washing than was the case with mechanical mixtures.

Addition of wetting agents did not seem to affect tenacity.

Addition of fungicides such as Bordeaux mixture and copper oxide likewise did not affect tenacity.

LITERATURE CITED

- 1. BLISS, C. I., 1935. The calculation of the dosage mortality curve. Ann. Appl. Biol. 22:135-67.
- 2. Busvine, J. R. and S. Barnes, 1947. Observations on mortality among insects exposed to dry insecticidal films. Bul. Ent. Res. 38:81-90.
- 3. Chisholm, R. D., 1945. Formulation of DDT sprays and dusts. U. S. D. A. Bur. Ent. & Pl. Quar. E643.
- 4. GARMAN, P., 1943. Control of the apple maggot with rotenone dusts. Conn. Agr. Expt. Sta. Bul. 474.
- 5. ———, 1945. A study of stickers for lead arsenate sprays on fruit trees. Conn. Agr. Expt. Sta. Bul. 485.
- 6. Gunther, F. A., D. L. Lindgren, M. I. Elliot and J. P. LaDue, 1946. Persistence of DDT deposits under field conditions. Jour. Econ. Ent. 39:624-627.
- 7. Heuberger, J. W., 1940. A laboratory biological assay of tenacity of fungicides. Phytopath. 30:840-847.
- 8. HORSFALL, J. G., 1945. Fungicides and Their Action. 239 pp. Chronica Botanica Co., Waltham, Mass.
- 9. Kennedy, J. S., 1947. The excitant and repellent effects on mosquitoes of sublethal contacts with DDT. Bul. Ent. Res. 38:593-607.
- 10. LINDQUIST, A. W. AND H. G. WILSON, 1948. Development of a strain of house flies resistant to DDT. Science 107:276.
- 11. MACINNES, D. G., 1947. On the effect of DDT-treated surfaces on adults of *Anopheles gambiae* and *A. funestris*. Bul. Ent. Res. 38:123-130.
- 12. Parkin, E. A. and A. A. Green, 1947. DDT residual films. 1. The persistence and toxicity of deposits from kerosene solutions on wall-board. Bul. Ent. Res. 38:311-325.
- 13. Skaptason, J. B. and F. M. Blodgett, 1941. Factorial studies on potato dusting materials. Amer. Pot. Jour. 18:1-9.
- 14. Turner, N., 1945. The coverage factor in the application of dusts. Jour. Econ. Ent. 38:359-365.
- 15. ——— AND N. WOODRUFF, 1946. Factors affecting the toxicity of DDT residues. In Conn. Agr. Expt. Sta. Bul. 512:93-97.
- 16. Woodruff, N. and N. Turner, 1947. The effect of particle size on the toxicity of DDT diluents in water suspension. Jour. Econ. Ent. 40:206-210.

